Pending Claims and Status

Claims 1-27 cancelled.

28. **[Currently Amended]** A method of producing a cross-over chemokine protein that contains at least one peptide segment whose sequence is derived from a first chemokine protein, and at least one peptide segment whose sequence is derived from a second chemokine protein, wherein said second chemokine protein has an amino acid sequence that is different from that of said first chemokine protein, and wherein each of said peptide segments possesses an N-terminal amino acid residue and a C-terminal amino acid residue, said method comprising:

ligating under chemoselective chemical ligation conditions (i) at least one peptide segment comprising a functional protein module derived from said first chemokine protein, and (ii) at least one peptide segment comprising a functional protein module derived from said second chemokine protein, wherein each of said peptide segments exhibit sufficient homology to a functional [domain] protein module of a chemokine[, macrophage migration inhibitory factor, cytokine, trefoil peptide, growth factor, protease inhibitor, or protein toxin,] to permit said peptide segments to mediate the function of [said] the chemokine functional [domain] protein module when incorporated into said cross-over chemokine protein, and wherein the C-terminal residue of said peptide segment derived from said first chemokine protein and the N-terminal residue of said peptide segment derived from said second chemokine protein comprise compatible reactive groups capable of chemoselective chemical ligation to one another, whereby a covalent bond is formed between said compatible reactive groups of said peptide segments so as to produce a chemical ligation product comprising a cross-over chemokine protein in which the C-terminal residue of the peptide segment derived from said first chemokine protein is ligated to the N-terminal residue of said peptide segment derived from said second chemokine protein[; wherein said first and second proteins are

jointly selected from the group consisting of chemokines, macrophage migration inhibitory factors, cytokines, trefoil peptides, growth factors, protease inhibitors, and protein toxins].

29. **[Currently Amended]** The method of claim 28 further comprising the step of conducting one or more additional ligations with one or more additional peptide segments, each possessing an N-terminal amino acid residue and a C-terminal amino acid residue, [wherein each of said one or more additional peptide segments exhibit sufficient homology to a functional domain of a chemokine, macrophage, migration inhibitory factor, cytokine, trefoil peptide, growth factor, protease inhibitor, or protein toxin, to permit said peptide segments to mediate the function of said functional domain when incorporated into said cross-over protein,]wherein said additional peptide segments [are selected from the group consisting of a peptide whose C-terminal residue comprises a reactive group capable of chemoselective chemical ligation with a reactive group of an N-terminal residue of another peptide, and a peptide whose N-terminal residue comprises] comprise a compatible reactive group capable of chemoselective chemical ligation to said cross-over chemokine protein [a reactive group capable of chemoselective chemical ligation with a reactive group of an C-terminal residue of another peptide].

30. **[Cancelled]**

31. **[Previously Presented]** The method of claim 28, wherein said chemoselective chemical ligation is selected from the group consisting of native chemical ligation, oxime forming chemical ligation, thioester forming ligation, thioether forming ligation, hydrazone forming ligation, thiazolidine forming ligation, and oxazolidine forming ligation.

32. **[Currently Amended]** A method of producing a cross-over chemokine protein library whose members contain two or more peptide segments, wherein each of said

peptide segments exhibit sufficient homology to a functional [domain] protein module of a chemokine [, macrophage migration inhibitory factor, cytokine, trefoil peptide, growth factor, protease inhibitor, or protein toxin,] to permit said peptide segments to mediate the function of said functional [domain] protein module when incorporated into said cross-over chemokine protein, each segment possessing an N-terminal amino acid residue and a C-terminal amino acid residue, and wherein the peptide segments of said members are derived from two or more different [proteins] chemokine protein molecules, said method comprising:

incubating under chemoselective ligation reaction conditions a plurality of unique peptide segments each comprising one or more functional protein modules derived from a member of a first set of chemokine protein molecules and a plurality of unique peptide segments each comprising one or more functional protein modules derived from a member of a second set of chemokine protein molecules wherein the C-terminal residues of each of said peptide segments derived from said members of said first set of protein molecules and the N-terminal residue of each of said peptide segments derived from said members of said second set of protein molecules comprise compatible reactive groups capable of chemoselective chemical ligation to one another, whereby a covalent bond is formed between said compatible reactive groups of said peptide segments so as to produce a plurality of chemical ligation products comprising a plurality of unique cross-over chemokine proteins, wherein, for each such cross-over chemokine protein, the C-terminal residue of a peptide segment derived from a member of said first set of chemokine protein molecules is ligated to the N-terminal residue of a peptide segment derived from a member of said second set of chemokine protein molecules[; wherein said first and second proteins are jointly selected from the group consisting of chemokines, macrophage migration inhibitory factors, cytokines, trefoil peptides, growth factors, protease inhibitors, and protein toxins].

33. **[Cancelled]**

34. **[Cancelled]**

35. **[Cancelled]**

36. **[Previously Presented]** The method of claim 32, wherein said chemoselective chemical ligation is selected from the group consisting of native chemical ligation, oxime forming chemical ligation, thioester forming ligation, thioether forming ligation, hydrazone forming ligation, thiazolidine forming ligation, and oxazolidine forming ligation.

Claims 37-51 cancelled.

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~~37.~~ **[New]** The method of claim 29, wherein said chemoselective chemical ligation is selected from the group consisting of native chemical ligation, oxime forming chemical ligation, thioester forming ligation, thioether forming ligation, hydrazone forming ligation, thiazolidine forming ligation, and oxazolidine forming ligation.

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~~38.~~ **[New]** The method of claim 29, further comprising the step of adding at least one chemical tag.

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~~39.~~ **[New]** The method of claim 38, wherein said chemical tags are the same or different and are independently selected from the group consisting of synthesis and purification handles, detectable labels, chemical moieties for attachment to a support matrix, and mixtures thereof.

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40. **[New]** The method of claim 38, wherein the chemical tag is selected from the group consisting of metal binding tags, carbohydrate/substrate binding tags, antibodies, antibody fragment tags, isotopic labels, haptens, unnatural amino acids, chromophores, and mixtures thereof.

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41. [New] The method of claim 38, further comprising the step of separating the chemical tag from the cross-over chemokine protein.

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42. [New] The method of claim 28 wherein the first and second chemokine proteins are derived from different subfamilies of chemokines.

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43. [New] The method of claim 28 wherein the first and second chemokine proteins are derived from different species.

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44. [New] The method of claim 28 further comprising the step of preparing the first and second chemokine proteins by chemical synthesis, ribosomomally in a cell free system, ribosomally within a cell, or any combination thereof.

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45. [New] The method of claim 28 wherein the first chemokine protein is chemically modified to incorporate D-amino acids, other unnatural amino acids, ester backbone bonds to replace the normal amide bond, alkyl backbone bonds to replace the normal amide bond, N- alkyl substituents, C- alkyl substituents, side chain modifications, disulfide bridges, side chain amide linkages, side chain ester linkages, and combinations thereof.

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46. [New] The method of claim 28 wherein the second chemokine protein is chemically modified to incorporate D-amino acids, other unnatural amino acids, ester backbone bonds to replace the normal amide bond, alkyl backbone bonds to replace the normal amide bond, N- alkyl substituents, C- alkyl substituents, side chain modifications, disulfide bridges, side chain amide linkages, side chain ester linkages, and combinations thereof.

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~~47.~~ [New] The method of claim 32, further comprising the step of adding at least one chemical tag.

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~~48.~~ [New] The method of claim ~~47,~~ wherein said chemical tags are the same or different and are independently selected from the group consisting of synthesis and purification handles, detectable labels, chemical moieties for attachment to a support matrix, and mixtures thereof.

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~~49.~~ [New] The method of claim ~~47,~~ wherein the chemical tag is selected from the group consisting of metal binding tags, carbohydrate/substrate binding tags, antibodies, antibody fragment tags, isotopic labels, haptens, unnatural amino acids, chromophores, and mixtures thereof.

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~~50.~~ [New] The method of claim ~~47,~~ further comprising the step of separating the chemical tag from the cross-over chemokine protein.

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~~51.~~ [New] The method of claim 32 wherein the first and second chemokine proteins are derived from different subfamilies of chemokines.

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~~52.~~ [New] The method of claim 32 wherein the first and second chemokine proteins are derived from different species.

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~~53.~~ [New] The method of claim 32 further comprising the step of preparing the first and second chemokine proteins by chemical synthesis, ribosomomally in a cell free system, ribosomally within a cell, or any combination thereof.

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~~54.~~ [New] The method of claim 32 wherein the first chemokine protein is chemically modified to incorporate D-amino acids, other unnatural amino acids, ester backbone bonds to replace the normal amide bond, alkyl backbone bonds to replace the normal amide bond, N- alkyl substituents, C- alkyl substituents, side chain modifications, disulfide bridges, side chain amide linkages, side chain ester linkages, and combinations thereof.

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~~55.~~ [New] The method of claim 32 wherein the second chemokine protein is chemically modified to incorporate D-amino acids, other unnatural amino acids, ester backbone bonds to replace the normal amide bond, alkyl backbone bonds to replace the normal amide bond, N- alkyl substituents, C- alkyl substituents, side chain modifications, disulfide bridges, side chain amide linkages, side chain ester linkages, and combinations thereof.

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~~56.~~ [New] A method of producing a cross-over chemokine protein comprising a single ligation site, comprising:

selecting a first peptide segment whose sequence is derived from a first chemokine protein wherein said first peptide segment possesses an N-terminal amino acid residue and a C-terminal amino acid residue and a functional protein module derived from said first chemokine protein, and wherein said first peptide segment exhibits sufficient homology to a functional protein module of a chemokine to permit said first peptide segment to mediate the function of the chemokine functional protein module when incorporated into said cross-over chemokine protein,

selecting a second peptide segment whose sequence is derived from a second chemokine protein, wherein said second peptide segment possesses an N-terminal amino acid residue and a C-terminal amino acid residue and a functional protein module derived from said second chemokine protein, and wherein said first peptide segment exhibits sufficient homology to a functional protein module of a chemokine to mediate the function of the

chemokine functional protein module when incorporated into said cross-over chemokine protein, and wherein said second chemokine protein has an amino acid sequence that is difference from that of said first chemokine protein,

ligating under chemoselective chemical ligation conditions said first peptide segment with said second peptide segment so that the C-terminal residue of said first peptide segment and the N-terminal residue of said second peptide segment undergo chemoselective chemical ligation to one another, whereby a covalent bond is formed between said compatible reactive groups of said first and second peptide segments so as to produce a chemical ligation product comprising a cross-over chemokine protein in which the C-terminal residue of the first peptide segment is ligated to the N-terminal residue of said second peptide segment.

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~~57.~~ [New] The method of claim ~~56,~~ further comprising the step of adding at least one chemical tag.

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~~58.~~ [New] The method of claim ~~57,~~ wherein said chemical tags are the same or different and are independently selected from the group consisting of synthesis and purification handles, detectable labels, chemical moieties for attachment to a support matrix, and mixtures thereof.

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~~59.~~ [New] The method of claim ~~57,~~ wherein the chemical tag is selected from the group consisting of metal binding tags, carbohydrate/substrate binding tags, antibodies, antibody fragment tags, isotopic labels, haptens, unnatural amino acids, chromophores, and mixtures thereof.

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~~60.~~ [New] The method of claim ~~57,~~ further comprising the step of separating the chemical tag from the cross-over chemokine protein.

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~~61.~~ **[New]** The method of claim ~~56~~ wherein the first and second chemokine protein segments are derived from different subfamilies of chemokines.

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~~62.~~ **[New]** The method of claim ~~56~~ wherein the first and second chemokine protein segments are derived from different species.

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~~63.~~ **[New]** The method of claim ~~56~~ further comprising the step of preparing the first and second chemokine protein segments by chemical synthesis, ribosomomally in a cell free system, ribosomally within a cell, or any combination thereof.

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~~64.~~ **[New]** The method of claim ~~56~~ wherein the first protein segment is chemically modified to incorporate D-amino acids, other unnatural amino acids, ester backbone bonds to replace the normal amide bond, alkyl backbone bonds to replace the normal amide bond, N-alkyl substituents, C- alkyl substituents, side chain modifications, disulfide bridges, side chain amide linkages, side chain ester linkages, and combinations thereof.

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~~65.~~ **[New]** The method of claim 28 wherein the second protein segment is chemically modified to incorporate D-amino acids, other unnatural amino acids, ester backbone bonds to replace the normal amide bond, alkyl backbone bonds to replace the normal amide bond, N- alkyl substituents, C- alkyl substituents, side chain modifications, disulfide bridges, side chain amide linkages, side chain ester linkages, and combinations thereof.

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~~66.~~ **[New]** A method of producing a cross-over chemokine protein, said method comprising:

ligating under chemoselective native chemical ligation conditions (i) a first peptide segment comprising an N-terminal cysteine and a functional protein module derived from a

first chemokine protein, and (ii) a second peptide segment comprising a C-terminal thioester and a functional protein module derived from a second chemokine protein, wherein said first and second chemokine proteins each have different amino acid sequences, whereby a native peptide bond is formed between said N-terminal cysteine and said C-terminal thioester so as to produce a chemical ligation product comprising said cross-over chemokine protein.